



Notman, Gillian M., McGill, Rona A.R., Hawkins, Stephen J. and Burrows, Michael T. (2016) Macroalgae contribute to the diet of *Patella vulgata* from contrasting conditions of latitude and wave exposure in the UK. *Marine Ecology Progress Series*, 549 . pp. 113-123.

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Macroalgae contribute to the diet of *Patella vulgata* from contrasting conditions of latitude
and wave exposure in the UK

Consumption of macroalgae by *Patella vulgata*

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Abstract

Analysis of gut contents and stable isotope composition of intertidal limpets (*Patella vulgata*) showed a major contribution of macroalgae to their diet, along with microalgae and invertebrates. Specimens were collected in areas with limited access to attached macroalgae, suggesting a major dietary component of drift algae. Gut contents of 480 animals from two moderately wave exposed and two sheltered rocky shores in each of two regions: western Scotland (55-56°N) and southwest England (50°N), were analysed in two years (n = 30 per site per year). The abundance of microalgae, macroalgae and invertebrates within the guts was quantified using categorical abundance scales. Gut content composition was compared among regions and wave exposure conditions; showing that the diet of *P. vulgata* changes with both wave exposure and latitude. Microalgae were most abundant in limpet gut contents in animals from southwest sites, whilst leathery/corticated macroalgae were more prevalent and abundant in limpets from sheltered and northern sites. *P. vulgata* appears to have a more flexible diet than previously appreciated and these keystone grazers consume not only microalgae, but also large quantities of macroalgae and small invertebrates. To date, limpet grazing studies have focussed on their role in controlling recruitment of macroalgae by feeding on microscopic propagules and germlings. Consumption of adult algae suggests *P. vulgata* may also directly control the biomass of attached macroalgae on the shore, whilst consumption of drift algae indicates the species may play important roles in coupling subtidal and intertidal production.

Introduction

The intertidal limpet, *Patella vulgata*, is the most abundant grazer on rocky shores in the north east Atlantic and plays an important role in structuring intertidal communities throughout its range (Hawkins & Hartnoll 1983, Hawkins et al. 1992). Classic limpet removal experiments (Jones 1948, Lodge 1948, Burrows & Lodge 1950, Southward 1964), observations following the mass limpet kills in the aftermath of the Torrey Canyon oil spill clean-up (Southward & Southward 1978, Hawkins & Southward 1992) and limpet exclusion experiments (Hawkins 1981, Hartnoll & Hawkins 1985, Jenkins et al. 2005, Coleman et al. 2006, Jonnson et al. 2006) have all shown that removal or exclusion of limpets produces lush growth of canopy forming furoid macroalgae on wave exposed and moderately exposed rocky shores. These indirect studies have led to the conclusion that *P. vulgata* is a keystone species in the intertidal, preventing macroalgal growth through microphagous grazing of epilithic biofilm and associated macroalgal propagules (spores and germlings) (Hill & Hawkins 1991, Jenkins & Hartnoll 2001, Jenkins et al. 2005, Coleman et al. 2006, Jonnson et al. 2006, Moore et al. 2007). This view is supported by observational studies of limpet foraging behaviour (Hartnoll & Wright 1977, Little et al. 1988, Della Santina et al. 1994) and several studies which directly examined limpet gut contents (Hawkins et al. 1989, Hill & Hawkins 1991, Little et al. 1990).

More recent work suggests that *P. vulgata* may play an even more significant role in structuring rocky shore ecosystems through direct grazing of mature macroalgae as well as biofilm and propagules. On many more sheltered shores *P. vulgata* aggregate around established stands of *Ascophyllum nodosum* and *Fucus vesiculosus* and ‘bite marks’ are visually apparent on the algal fronds. The fronds are often much shorter than might be expected, consistent with the ends being directly grazed away by the nearby limpets (Davies et al. 2007, Davies et al. 2008, personal observation). Lorenzen (2007) also recorded *P.*

vulgata feeding on stranded macroalgae during periods of emersion, and the results of stable isotope analyses from several sources provide strong evidence to suggest that macroalgae may be a more significant source of organic carbon to *P. vulgata* than previously appreciated (Campbell 2004, Riera et al. 2009, Schaal et al. 2010, Notman 2011).

The limited number of studies which have examined gut contents in *P. vulgata* to date probably give an incomplete view of the dietary range of *P. vulgata*. Sample sizes for foregut analysis were small and acid digestion methods may have provided biased information on only a subset of the diet rather than representing whole gut contents (Hawkins et al. 1989, Hill & Hawkins 1991). This has led to an overemphasis on grazing of the microalgal biofilm by the species, and a potential underestimation of their capacity for feeding on adult or detrital macroalgae. Quantification of whole diet composition without such bias is lacking, and is crucial for a re-evaluation of the role of the species as a keystone grazer across its range.

The overall aim of this study was to compare the diets of limpets between contrasting latitudinal and wave exposure conditions. Specifically we quantified the relative importance of microalgal biofilms and other food sources across these locations. We focussed on the diet of limpets collected from bare rock microhabitats where access to other types of foods was restricted. Gut contents of adult *P. vulgata* (30-50 mm in length) were examined from wave exposed and wave sheltered sites in western Scotland and southwest England over two years. Analysis of carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) stable isotope ratios from a subset of these animals, together with their potential foods, was used to provide additional information on limpet diets integrated over longer periods of time.

Materials and Methods

Study sites & sample collection

Patella vulgata were collected from mid-shore bare rock microhabitats at four sites in southwest England, near Plymouth, Devon, and four sites in western Scotland, around Oban and the Kintyre peninsula. Two moderately wave-exposed (300 – 500 km fetch) and two sheltered (< 50 km fetch) sites were selected in each region, with the ranges of wave exposure set using calculated values for wave fetch in accordance with Burrows *et al.* (2008). Adult limpets were collected from areas of bare rock in spring 2005 and 2006. These habitats seldom comprised entirely clean rock, but also often contained small areas of macroalgae and barnacles. To investigate the potential influence of these alternate food sources on diet composition, we visually estimated the percentage cover of bare rock, barnacle and attached macroalgal cover in the mid-intertidal using twenty five 0.5 × 0.5 m quadrats at the eight sites in each year. Limpets were immediately chilled upon collection to minimise digestion of gut contents, and frozen (-20°C) on return to the laboratory. Thirty animals of similar size (mean length 42 ± 4 mm) were selected from each site per year for gut contents analysis. A subset of twelve limpets were randomly selected for stable isotope analysis from the different locations and wave exposure conditions, and samples of potential foods (biofilm and macroalgae) were collected at each site in each year. Macroalgal species included *Ascophyllum nodosum*, *Fucus vesiculosus*, *F. serratus*, *F. spiralis*, *Himanthalia elongata*, and detrital *L. digitata*.

Dissection of gut contents

The digestive organs of *P. vulgata* are long and complex, extending from the mouth and buccal mass, through the crop and stomach to the long intestine embedded in the visceral mass (Fretter & Graham, 1962). Limpets were dissected ventrally, whilst partially frozen, with shells attached. The muscular foot and internal organs were completely removed to

110 reveal the crop and anterior portion of the stomach. Only the anterior sections of the
111 alimentary canal were excised as the posterior sections are difficult to separate from the
112 digestive gland and visceral mass.

113 **Examination of gut contents**

114 Dissected gut contents were initially examined at $\times 4$ magnification, and ingested algae
115 and invertebrates identified and quantified using categorical abundance scales (Table 1). It
116 was not possible to identify ingested macroalgae to species level. These were therefore
117 classified according to Steneck and Watling's (1982) functional group scheme (Table 1), with
118 categories 4 and 5 (corticated and leathery macroalgae) combined due to the difficulties in
119 objectively and consistently discriminating between these groups in gastropod guts (Raffaelli,
120 1985, personal observation). The abundance of microalgae (Group 1 including diatoms and
121 cyanobacteria) was assessed at $\times 20$ magnification using a compound microscope. A 1 cm²
122 subsample of material was covered with a glass coverslip and five fields of view were
123 randomly selected for closer examination.

124 **Stable isotope analysis**

125 Tissue from the muscular foot of *P. vulgata* was chosen for isotope analysis because
126 muscle tissues integrate consumer diets over long periods of time (Hobson & Clark 1992)
127 and, due to their low fat content, do not require lipid extraction (Pinnegar & Polunin 1999).
128 Dissected foot tissues from 48 of the limpets used in gut contents analysis were rinsed in
129 deionised water and freeze dried to constant weight for stable isotope analysis.

130 Biofilm was obtained from three 200 cm² rock samples (free from macroalgae and
131 encrusting organisms) from each site in each year. The rocks were soaked in 0.7 μ m filtered
132 seawater for 15 minutes in the laboratory and the rehydrated biofilm removed by brushing
133 with an electric toothbrush. Biofilm was filtered onto glass fibre filter paper (Whatman
134 GF/F), frozen at -20°C and freeze dried to constant weight. Samples of corticated/leathery

macroalgae were rinsed in deionised water and epibiont-free fronds were frozen at -20°C before freeze drying. The stable isotope values of barnacles were not measured due to the difficulties inherent in analysing inorganic carbon (Soreide *et al* 2006).

Samples were ground into homogenous powders, weighed into tin capsules (0.7 mg limpet tissue, 1.5 mg macroalgal tissue and two 9 mm discs of biofilm/filter paper), randomised and loaded into an automatic carousel for simultaneous analysis of carbon and nitrogen isotopes using continuous-flow isotope ratio mass spectrometry (CF-IRMS) (Costech model ECS 4010 elemental analyser coupled with a ThermoFinnegan Delta Plus XP mass spectrometer). Two laboratory standards were analysed every 10 samples allowing instrument drift to be corrected if required.

Data analysis

Abundance of algae and invertebrates ingested by limpets was compared between the two regions, between the two classes of wave exposure and among the sites (nested within region and exposure), using ordinal logistic regression (OLR, Minitab version 15.1.20, MINITAB Inc.) on ranked categorical abundance data to determine influences on the probability of obtaining particular abundance categories. Best OLR models were selected by comparisons of deviance likelihood ratios following step-wise exclusion of factors from a saturated model (Notman 2011).

Multivariate analyses were used to examine patterns of gut content composition between the sampling regions and wave exposure conditions. The Gower similarity coefficient (S_{15}) was used to obtain a matrix of similarities for examination of ordinal categories of limpet gut contents (Gower, 1971; Podani, 1999; Legendre & Legendre, 1998). Normalisation was not necessary as similar abundance scales were used for each variable. Non-metric multidimensional scaling (MDS, PRIMER 6, PRIMER-E Ltd.) was used to produce a two-dimensional ordination of gut contents data from the similarity matrix (Clarke,

1993). Similarity indices were used to determine the effects of region and wave exposure on composition of limpet gut contents (2-way ANOSIM), with the contribution of each taxon to differences among levels of each factor assessed using similarity percentage analysis (SIMPER, based on the Euclidian distance measure of association).

Consumption of small invertebrates was assessed using counts of ingested items in a hierarchical cluster analysis. A matrix of similarities of ingested fauna was calculated for each site using the Bray-Curtis similarity coefficient following square root transformation of ingested faunal abundance data (Bray & Curtis, 1957) and analyses of similarity (ANOSIM) were used to assess whether the observed patterns of ingested invertebrates differed significantly between sampling regions and wave exposure conditions.

Stable carbon and nitrogen isotope ratios of biofilm, macroalgae and *P. vulgata* foot tissue were compared among the two regions, the two classes of wave exposure and the sites (nested within region and exposure) using analysis of variance. Site was treated as a random factor, with all other factors fixed and orthogonal. Stable isotope ratios for macroalgae were calculated for region and exposure type combinations as the mean and standard deviation across all species of corticated/leathery macroalgae for each site.

Habitat composition was evaluated using Mann-Whitney U-tests to compare percentage cover of bare rock at sampling sites among regions and Wilcoxon signed ranks tests to evaluate bare rock cover among levels of wave exposure within each region (with Bonferroni corrections for multiple comparisons) (SPSS version 22.0, IBM Corp).

Results

Study Sites

The percentage cover of bare rock was similar between moderately exposed sites in both regions at 92 % (Mann-Whitney U-test, $p = 0.75$). Although bare microhabitats were deliberately chosen at all sites, the percentage bare rock was lower on sheltered shores, around 88 % in the north and 84 % in the south, and differed significantly between regions (Mann-Whitney U-test, $p < 0.01$). Percentage macroalgal and barnacle cover was highest at southern sheltered shores at around 3 and 13 % respectively ($p < 0.01$ for all comparisons).

Main constituents of limpet gut contents

Most of the 480 animals examined had quantifiable gut contents (82 %). It was not possible to identify the species of algae ingested, but microalgae (Group 1) and corticated/leathery algae (Groups 4/5) were present in almost all of these samples: 90 % and 95 % of gut contents respectively. Filamentous (Group 2) algae were found in around 36 % of gut samples, and foliose (Group 3), articulated calcareous (Group 6) and crustose coralline (Group 7) algae were only present in around 10 % of the material analysed (Fig. 1). Invertebrates were present in 91 of the 392 limpet gut contents (23 % overall).

Changes in diet with latitude

There were significant differences in limpet diets between the two sampling regions. Microalgae were almost five times more likely to be recorded in high abundance categories (common, abundant and superabundant) in gut contents of southern limpets than northern limpets (ordinal logistic regression region effect, log odds ratio 1.514, SE 0.367, $p < 0.001$, Table 2, Fig. 2). In contrast, corticated/leathery macroalgae were around twice as likely to be recorded in higher abundance categories in northern limpets (Fig. 3). The odds ratio of 0.55 given in Table 2 indicates that high abundance categories of corticated/leathery algae were around half as likely in the south than in the north ($p < 0.01$). Abundance of other types of

algae (filamentous, foliose, articulated calcareous or crustose coralline) was not influenced by latitude ($p > 0.05$ for all comparisons).

Effect of wave exposure on diet

Microalgae were more prevalent in guts of limpets from exposed shores than sheltered shores in the south (Fig. 2) but not in the north, and no overall trend in the abundance of microalgae with wave exposure was seen. In the south, higher abundance categories for microalgae were more likely at the exposed shores at Andurn and Picklecombe (sites SE1 and SE2) than at the sheltered sites Jennycliff or Cawsand (SS1 or SS2; $p < 0.01$, Fig. 2, Table 2). There were no significant differences in microalgal abundance among the sites in the north, but there was some evidence that the probability of higher microalgal abundances was greater in samples from the moderately exposed Easdale (NE1) than for those from the nearby, sheltered Ellenabeich (NS1; odds ratio = 1.95, $p = 0.079$, Table 2).

Corticated/leathery algae were almost twice as likely to be at least common (Table 1) in the gut contents of limpets from sheltered sites than in those from exposed sites (odds ratio = 1.96, $p < 0.001$, Table 2 and Fig. 3). Abundance categories of filamentous, foliose, articulated calcareous or crustose coralline algae were not significantly affected by wave exposure ($p > 0.05$ for all comparisons).

Ingestion of invertebrates

Small invertebrates were present in 23 % of quantifiable limpet gut contents and were found at every site (9 to 42 % of limpets examined). Barnacles were the most frequently ingested taxon, found in 15 % of limpet guts. The small snail *Skeneopsis planorbis* was ingested by 6 % of limpets, and acarid mites were found in 3% of guts. Other taxa included bivalves (present in 4 individuals), foraminifera (3), ostracods (3), *Littorina mariae* (2), copepods (1), hydroids (1), *Melaraphe neritoides* (1), and *Patella vulgata* itself (1). Few limpets had ingested more than one type of invertebrate (only 4 %). Hierarchical cluster

analysis of ingested invertebrates and analyses of similarity indicated that there were no significant effects of sampling region or wave exposure on the faunal composition of limpet gut contents (ANOSIM region effect $r = -0.25$, $p = 0.78$; ANOSIM wave exposure effect $r = -0.25$, $p = 1.0$).

Patterns of gut content composition in *P. vulgata*

Multidimensional ordination of the abundance of the functional groups of algae and ingested fauna (Fig. 4) also showed that the composition of the diet of *P. vulgata* varied with region and wave exposure (ANOSIM region effect $r = 0.017$, $p = 0.025$; ANOSIM wave exposure effect $r = 0.03$, $p = 0.001$). Although no clear, discrete groupings of diet types are immediately apparent from the ordination plot, there is a general pattern for individuals from exposed sites in the south to be positioned towards the bottom right of the figure, and individuals from sheltered sites in both regions to be positioned towards the top left. Samples from northern exposed sites show less pronounced separation than other samples and overlap considerably with other data. The MDS plot (Fig. 4) places individual limpets with high abundance categories for microalgae (Fig. 4c) towards the right of the figure, and shows them to be mainly from exposed and southern sites. Individuals with high abundance categories for corticated/leathery algae (Fig. 4f) are positioned towards the top left of the MDS plot and tend to be from sheltered sites in the north. Limpets which had ingested invertebrate fauna are positioned towards the bottom of the ordination (Fig. 4b), and show no clear patterns according to wave exposure conditions or sampling region. The few individuals with high abundance categories for filamentous, foliose, articulated calcareous and encrusting coralline algae (Groups 2, 3, 6 and 7; Figs. 4d, e, g and h respectively) appear as outliers in the MDS plot and show no significant patterns according to the factors examined.

SIMPER analyses indicate that microalgae, corticated/leathery and filamentous algae contribute most to differences in diet composition by region and wave exposure (Table 3).

Average abundance categories for microalgae and filamentous algae were higher for samples from southern sites and average abundance categories for corticated/leathery algae were higher for limpet gut contents from northern and sheltered sites. Higher abundance categories of microalgae (Group 1 algae) were found in samples from wave exposed sites than in those from sheltered areas.

Stable isotopes

Analysis of variance of the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in *P. vulgata* foot tissues by region, wave exposure and site (nested within region and exposure) showed a significant effect of region on carbon isotope composition and significant effects of site on both carbon and nitrogen isotope ratios (Table 4, Fig. 5). The average $\delta^{13}\text{C}$ values of animals from the south were around 2 ‰ enriched in ^{13}C compared to animals from the north ($p < 0.01$). Variability in $\delta^{15}\text{N}$ values of limpet tissues was largely due to differences between sites ($p < 0.01$) and, although nitrogen isotope values in limpets from exposed sites in the north were slightly higher than those of sheltered sites in this region, this trend was not significant ($p > 0.05$). Average $\delta^{13}\text{C}$ values for corticated/leathery macroalgae differed significantly between the sampling regions (-16.4 ‰ in the north and -17.2 to -18.5 ‰ in the south; $p < 0.01$; Fig. 5), and average $\delta^{15}\text{N}$ values were lower in the north (5.3 to 6.2 ‰) compared to similar algae from the south (7.4 to 7.6 ‰). No significant wave exposure effects were seen. The average $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of epilithic biofilm were -20.7 ‰ and 6.4 ‰ respectively and no significant differences among region and wave exposure conditions were shown (Fig. 5).

Discussion

Our work shows that the intertidal limpet *Patella vulgata* consumes not only microalgae, but also large quantities of macroalgae and some small invertebrates. Intertidal consumers are thought to exploit the most common food sources available in their immediate surroundings (Raffaelli 1985; Steinarsdottir et al. 1995) and *P. vulgata* is a homing species which tends to forage within 1 m of its 'home scar' (Hartnoll & Wright 1977, Della Santina et al. 1995). Despite being collected from bare rock habitats, 95 % of the limpets examined in this study had macroalgae in their guts, and the stable isotope evidence indicates that assimilated carbon was from macroalgal sources. It is clear that these animals are exploiting a macroalgal food resource that is not immediately apparent on the shore at low tide. *Patella vulgata* has been observed feeding on drift algae stranded on the shore when exposed at low water, particularly on dark, damp days and following storms (Lorenzen 2007, GMN, SJH personal observations). South African limpets (*Patella argenvillei* and *P. granatina*) have been documented capturing and feeding on drifting kelp directly from the water column (Bustamante et al. 1995). Our findings suggest that *P. vulgata* may also be able to exploit drift algae from the water column when the tide is in.

The relative abundance of ingested algal material in limpet guts is affected by both latitude and wave exposure; microalgae are more common in the diet of limpets from southern and moderately wave exposed sites, whereas corticated/leathery macroalgae are more frequently encountered in animals from northern and sheltered coastlines. Stable isotope analyses show that foot tissues from limpets from all sites are enriched in ^{13}C relative to biofilm, also indicating that the organic carbon assimilated over time is largely derived from macroalgal sources rather than microalgae.

Patella vulgata plays a major role in structuring intertidal communities along gradients of wave exposure and latitude in the north-east Atlantic by ingestion of biofilm and

305 macroalgal propagules (Hawkins & Hartnoll 1983, Hawkins et al. 1992) and previous gut
306 contents analyses have supported this (Hawkins et al. 1989, Hill & Hawkins 1991). Ingestion
307 of invertebrates was reported in these studies at similar levels to those recorded here, but acid
308 digestion probably removed much of the macroalgal tissues from the analyses and the
309 presence of any residual macroalgal tissue (*Fucus* spp.) within the gut was interpreted as
310 consumption of juvenile algae, rather than sections of mature thalli (Hill & Hawkins 1991).
311 It is difficult dissect the limpet alimentary tract, and even harder to identify fragments of
312 algae in limpet gut contents (Raffaelli 1985, C. Maggs personal communication). Yet our
313 examination of gut contents, combined with stable carbon and nitrogen isotope analysis,
314 provides very strong evidence that *P. vulgata* routinely consumes mature macroalgae as well
315 as biofilm and macroalgal propagules. Use of live and detrital macroalgal foods may explain
316 the lack of clear relationships between biofilm standing stock, limpet density and grazing
317 activity which have been observed on a number of British shores (Jenkins et al. 2001,
318 Thompson et al. 2004).

319 Some of the ingested macroalgae in this study may be from *P. vulgata* directly
320 consuming adult furoid plants. In sheltered sea lochs and bays, where intertidal communities
321 are dominated by leathery, furoid macroalgae, *P. vulgata* may be directly feeding on the
322 fronds. On more exposed areas, headlands and breakwaters, where such algae are rare and
323 communities are characterised by barnacles and mussels (Lewis, 1964; Ballantine, 1961),
324 drift algae may enhance populations of these grazers and so maintain high grazing intensity
325 and prevent further escapes of furoids (Moore et al 2007). The greater availability of drift
326 algae in the north may explain the greater incidence of macroalgae in the diet of *P. vulgata*,
327 given that macroalgal cover increases with latitude. Furoids dominate most shores in
328 Norway whereas in northern Spain the algae are confined to only the most sheltered sites
329 (Ballantine 1961, Hawkins & Hartnoll 1983, Hawkins et al. 1992, Coleman et al. 2006).

By feeding on allochthonous drift algae, *P. vulgata* may be coupling subtidal and intertidal production through feeding. Thus their ecological role may be much more extensive than simply that of a microphagous grazer of rocky substrata. Such grazers may therefore play crucial roles in facilitating nutrient flow in coastal environments and allowing horizontal transport of resources between exposed and sheltered areas (Polis et al 1997).

Consumption of corticated/leathery algae is reported in many of the early accounts of limpet biology (see Steneck and Watling (1982) for a comprehensive review), and is also described in several more recent investigations (Davies et al., 2008; Davies et al., 2007; Lorenzen, 2007). Lorenzen (2007) provides photographic evidence of rasping marks made by *P. vulgata* on the thalli of *A. nodosum* from shores in Brittany and a video showing a low shore individual feeding on attached *Laminaria digitata* was recorded by G. Notman in Argyll in 2007 (<http://www.youtube.com/watch?v=79RvGRUdnwE>). Stable isotope evidence from this and other studies also indicates that fucoid macroalgae are a significant source of nutrition to *P. vulgata*; the limpets are not only ingesting macroalgal foods, but they are also assimilating them into their tissues (Campbell, 2004; Notman et al. in preparation; Riera et al., 2009; Schaal et al., 2010).

Despite limiting our investigation to relatively large limpets collected from open rock habitats, and acknowledging that the diet of these molluscs may change seasonally and ontogenetically, as well as in response to food availability, it is clear that the diet and the ecological role of *P. vulgata* is not yet fully understood. *Patella vulgata* is considered to be a keystone species on temperate rocky shores because its grazing activities prevent establishment of mature algae by consumption of macroalgal propagules and germlings in the biofilm. The assumption was that biofilm foods provided the main source of energy to these animals. Our work suggests that the species may also play important roles in modifying macroalgal cover, especially on more sheltered coastlines where fucoid algae commonly

355 occur. Moreover, previous work has interpreted the aggregation of *P. vulgata* under stands of
356 mature algae as being primarily a sheltering response, mitigating desiccation stress and
357 reducing predation (Hartnoll & Hawkins 1985, Coleman et al. 1999, Moore et al. 2007). This
358 work indicates that limpets associated with patches of attached macroalgae are likely to be
359 feeding on them too. The species may also be of great ecological importance in terms of
360 coupling sub- and intertidal production by ingesting allochthonous drift algae across a range
361 of wave exposure conditions and latitudes. This would help to explain the high biomass of
362 grazers which occurs on apparently bare shores across the north east Atlantic, which is
363 unlikely to be supported by epilithic microalgal production alone.

364

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483 Thompson RC, Roberts MF, Norton TA, Hawkins SJ (2000) Feast or famine for intertidal
484 grazing molluscs: A mis-match between seasonal variations in grazing intensity and the
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486 Table 1. *Patella vulgata* gut contents. Taxa identified and abundance categories used for gut contents analysis. Algal functional groups follow the
 487 classification by Steneck and Watling (1982).
 488

Group	Algal Type /Taxon	Representatives	Measure	S Super- abundant	A Abundant	C Common	F Frequent	O Occasional	R Rare	N None
1	Microalgae	Diatoms, cyanobacteria	No. cells per field of view	> 50	21 to 50	11 to 20	6 to 10	1 to 5	0.5 to 1	< 0.2
2	Filamentous Algae	<i>Cladophora</i> , <i>Ectocarpus</i>	No. of fragments	> 50	31 to 50	21 to 30	11 to 20	6 to 10	1 to 5	None
3	Foliose Algae	<i>Ulva</i> , <i>Porphyra</i>	No. of fragments	> 50	31 to 50	21 to 30	11 to 20	6 to 10	1 to 5	None
4	Corticated Macrophytes	<i>Bryothamnium</i> , <i>Chondria</i> /	No. of fragments	> 50	31 to 50	21 to 30	11 to 20	6 to 10	1 to 5	None
5	Leathery Macrophytes	<i>Laminaria</i> , <i>Fucus</i> , <i>Ascophyllum</i>	No. of fragments	> 50	31 to 50	21 to 30	11 to 20	6 to 10	1 to 5	None
6	Articulated Calcareous Algae	<i>Corallina</i> , <i>Halimeda</i>	No. of fragments	> 50	31 to 50	21 to 30	11 to 20	6 to 10	1 to 5	None
7	Crustose Coralline Algae	Lithothamnion, <i>Lithophyllum</i>	No. of fragments	> 50	31 to 50	21 to 30	11 to 20	6 to 10	1 to 5	None
Fauna	Ingested Invertebrates	Barnacle cyprids, <i>Skeneopsis planorbis</i> , acariniids	No. of individuals	> 50	31 to 50	21 to 30	11 to 20	6 to 10	1 to 5	None

Table 2. *Patella vulgata* gut contents. Comparison of abundance of microalgae and corticated/leathery macroalgae across regions, sites and wave exposure by ordinal logistic regression. Site type abbreviations: NE, northern exposed; NS, northern sheltered; SE, southern exposed; SS, southern sheltered. (***p < 0.001; **p < 0.01). Results are shown for best fit models (see text).

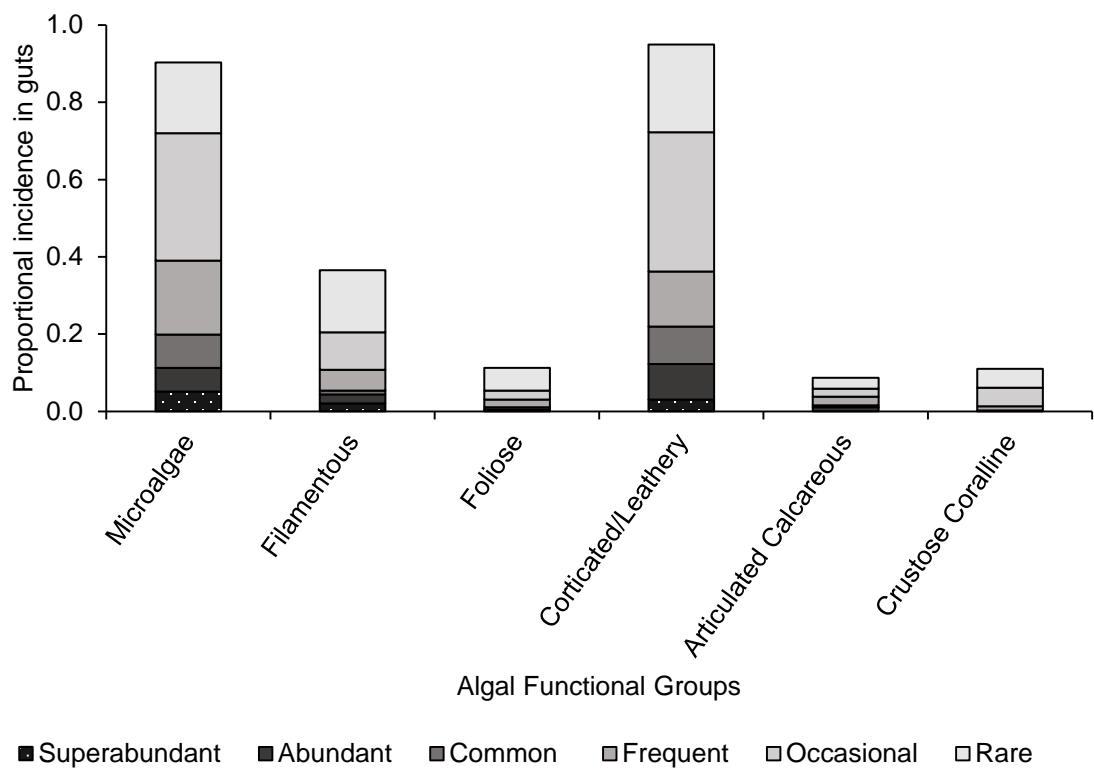
Region or Site code (region)	Loge odds ratio	SE	z	P	Odds ratio	95 % confidence interval upper lower	
<i>Microalgae: Group 1</i>							
North vs South	1.514	0.367	4.13	< 0.001***	4.54	2.22	9.32
NE1 Easdale							
NE2 Putechan	0.016	0.400	0.04	0.968	1.02	0.46	2.23
NS1 Ellenabeich	0.666	0.379	1.75	0.079	1.95	0.93	4.09
NS2 Luing	0.514	0.369	1.39	0.164	1.67	0.81	3.45
SE1 Andurn							
SE2 Picklecombe	0.429	0.348	1.23	0.217	1.54	0.78	3.04
SS1 Jennycliff	1.150	0.345	3.34	0.001**	3.16	1.61	6.21
SS2 Cawsand	1.654	0.347	4.77	< 0.001***	5.23	2.65	10.32
<i>Corticated/leathery algae: Group 4/5</i>							
North vs South	-0.606	0.184	-3.29	0.001**	0.55	0.38	0.78
Exposed vs Sheltered	0.672	0.184	3.64	< 0.001***	1.96	1.36	2.81

Table 3. *Patella vulgata* diet composition. Comparison of gut content composition between regions and wave exposures by SIMPER analysis using the Euclidian distance measure of association. Average abundance categories of algae are shown (categories N to S converted to numerical integers 0 to 7) with average squared Euclidian distance (D) and percentage contribution (C %) to differences between regions and wave exposures.

		Region				Wave exposure			
	Algal Group	North	South	D	C %	Exposed	Sheltered	D	C %
1	Microalgae	1.90	2.76	4.65	29	2.75	2.04	4.75	29
4/5	Corticated/Leathery	2.64	2.22	4.17	26	2.09	2.69	4.32	26
2	Filamentous	0.608	0.944	3.76	24	0.643	0.928	3.72	23
7	Encrusting coralline	0.08	0.315	1.11	7	0.346	0.087	1.34	8

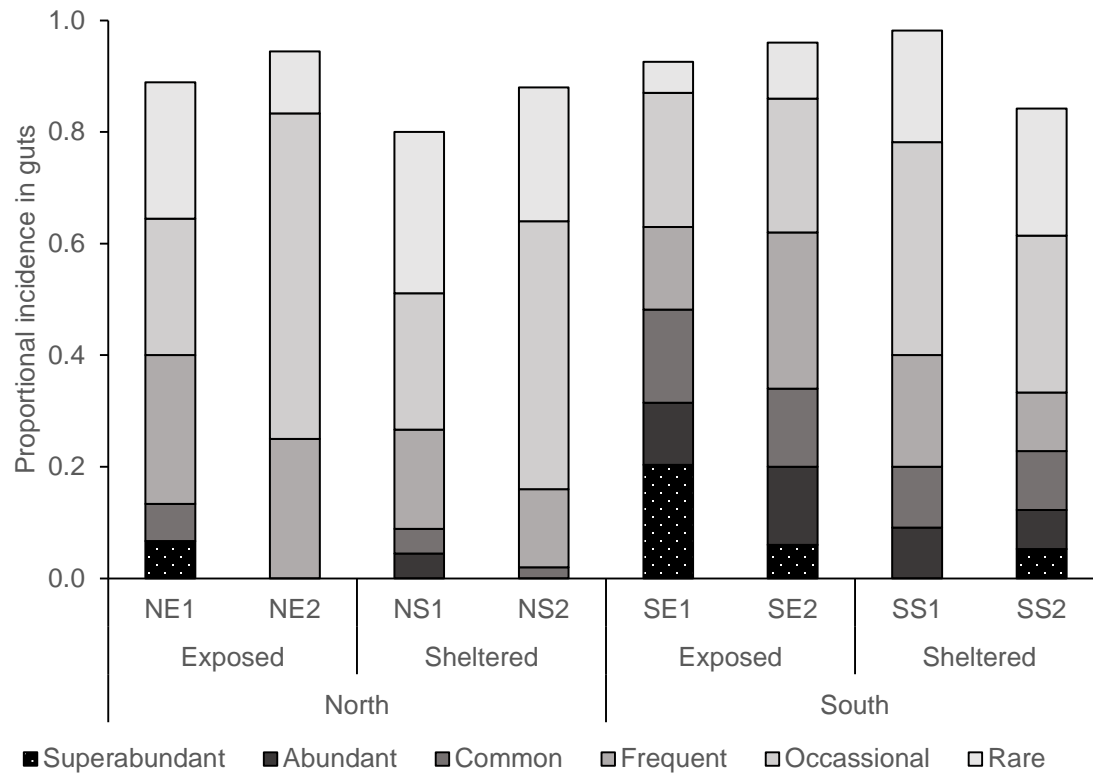
Table 4. ANOVA testing for differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values of *P. vulgata* foot tissue by region, wave exposure and site (nested within region and exposure). x denotes that the analysis could not perform an exact F test. (** $p < 0.01$) $n = 12$ per site.

Source	df	$\delta^{13}\text{C}$				$\delta^{15}\text{N}$			
		MS	F	p		MS	F	p	
Region	1	57.602	24.83	0.008**	x	7.157	4.28	0.107	x
Exposure	1	1.172	0.51	0.517	x	3.934	2.36	0.200	x
Site (Reg Exp)	4	2.323	5.89	0.001**		1.674	4.47	0.004**	
Region*Exposure	1	0.029	0.01	0.917	x	10.649	6.37	0.065	x
Error	44	0.394				0.374			
Levene's Test		1.22, $p = 0.315$				0.94, $p = 0.491$			
Bartlett's Test		5.89, $p = 0.553$				10.52, $p = 0.161$			
Variance Homogeneous		Yes				Yes			



514

515 Fig. 1. *Patella vulgata* gut contents (n = 392). Abundance of functional groups of algae in
516 gut contents of *P. vulgata* shown by cumulative proportional incidence in abundance
517 categories (defined in Table 1).



518

519 Fig. 2. *Patella vulgata* gut contents (n = 392). Abundance of microalgae by proportional
 520 incidence in abundance categories (Table 1) by region, wave exposure and site. Site
 521 abbreviations are given in Table 2.

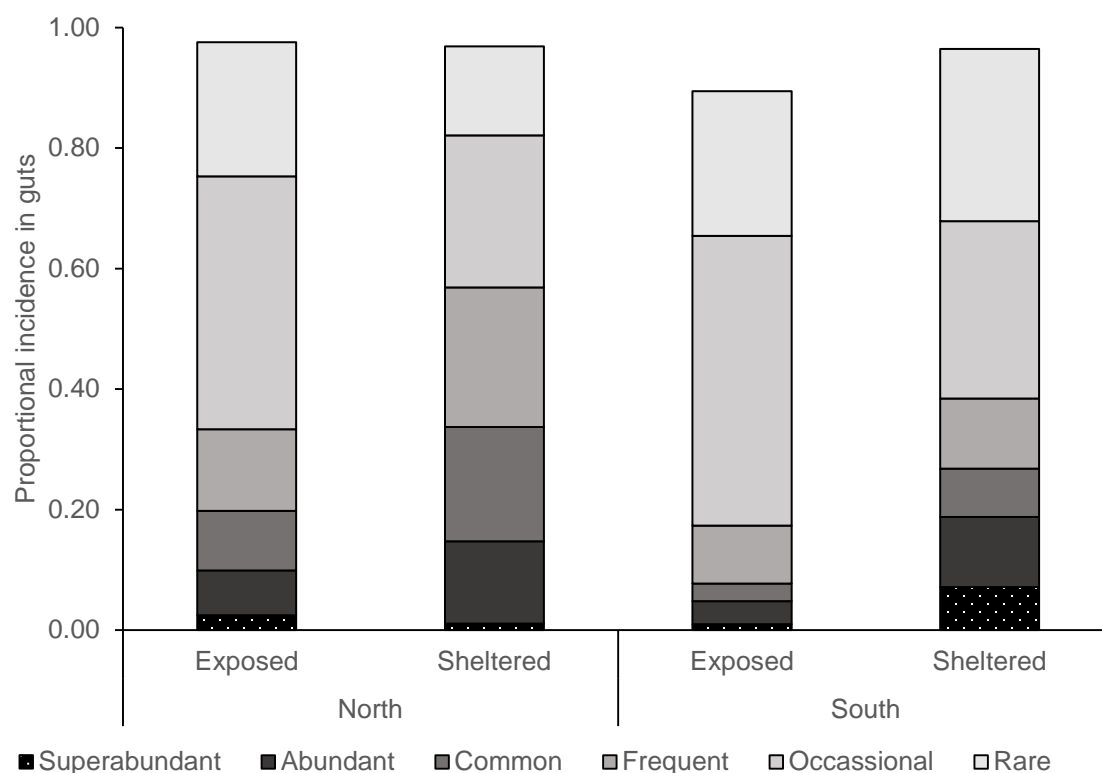


Fig. 3. *Patella vulgata* gut contents (n = 392). Abundance of corticated/leathery macroalgae as the proportional incidence in abundance categories (Table 1) by region and wave exposure.

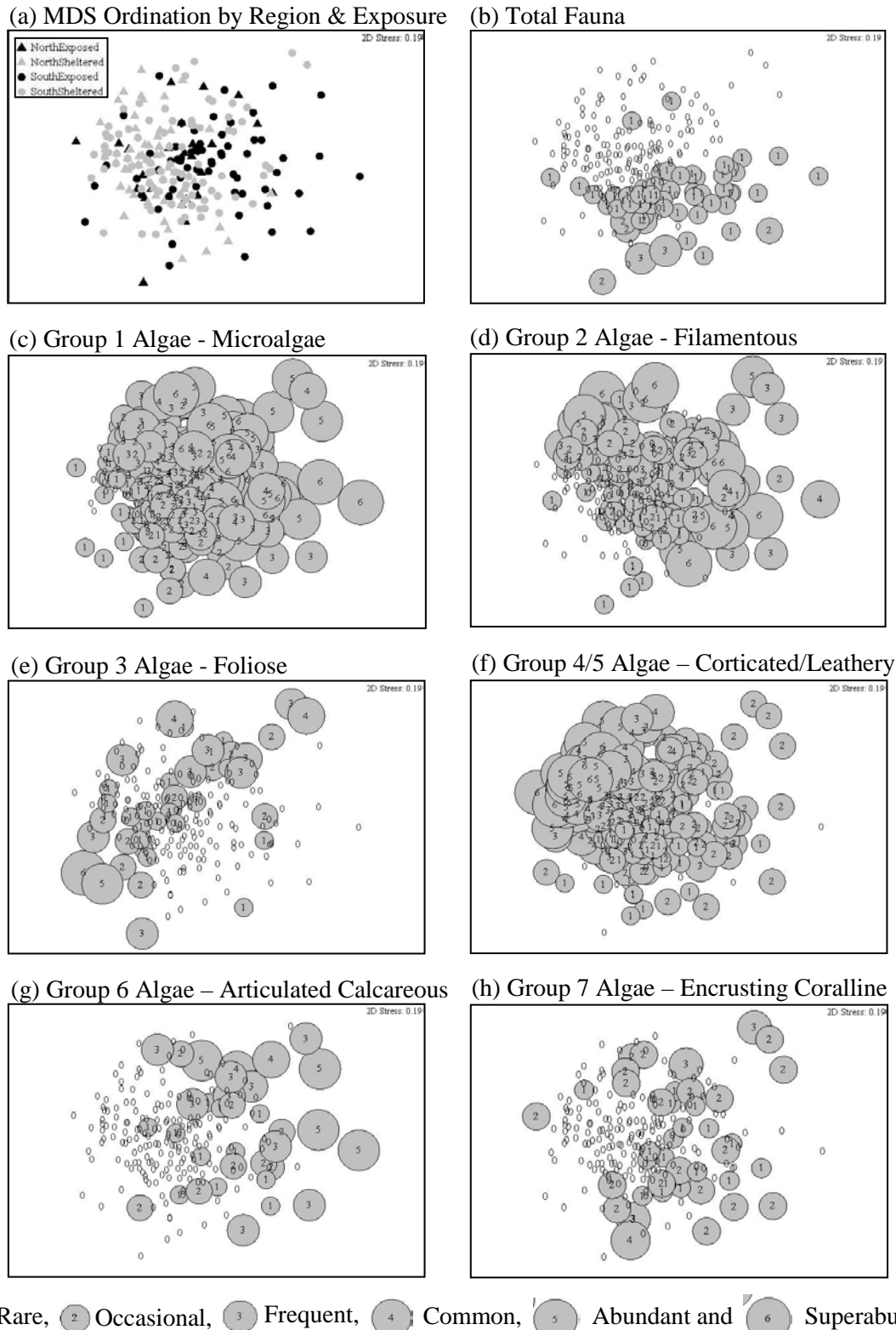


Fig. 4. *P. vulgata* gut contents. MDS ordination of the composition of individual *P. vulgata* diets using abundance categories converted to numerical integers using Gower's S_{15} similarity matrix ($n = 392$). Subplots show (a) region and wave exposure of collection sites (b) the abundance category for total fauna as a varying sized symbol, (c) microalgae, (d) filamentous algae, (e) foliose algae, (f) corticated and leathery algae, (g) articulated calcareous algae and (h) encrusting coralline algae.

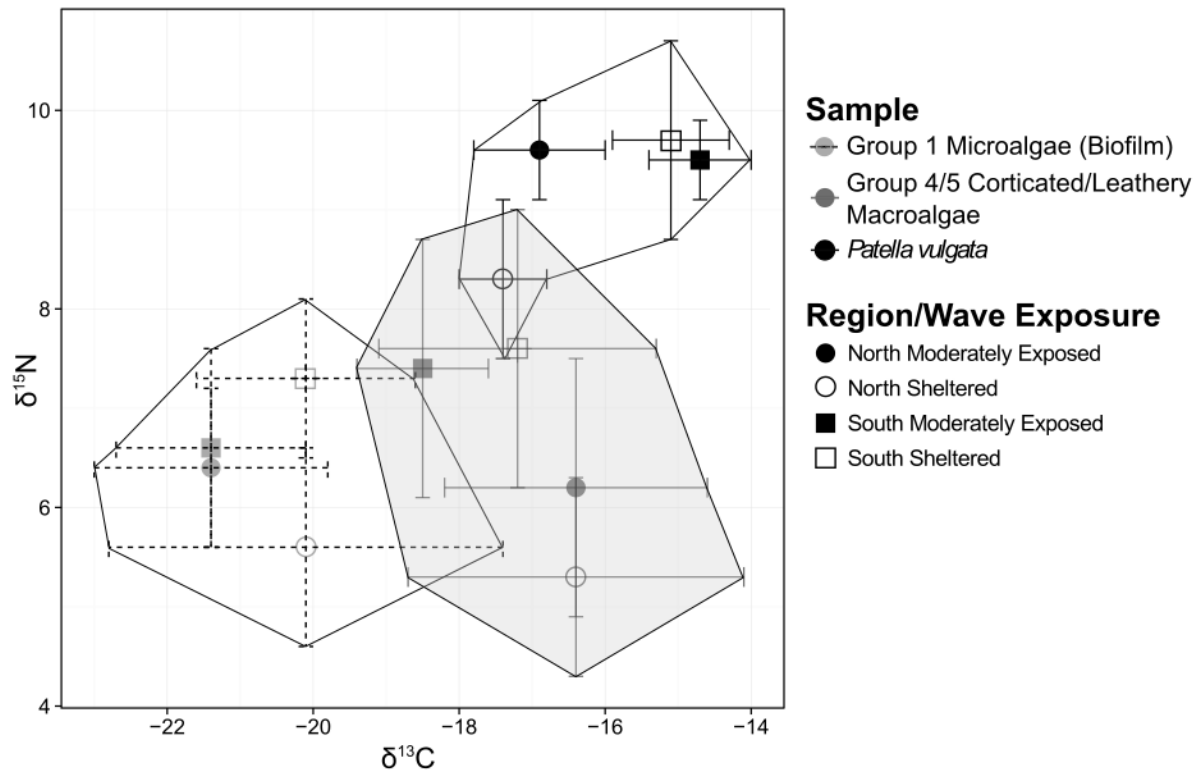


Fig. 5. Stable isotope ratios of Microalgae, Corticated/Leathery Macroalgae and *P. vulgata* foot tissues. Mean $\delta^{13}\text{C}$ values are given against mean $\delta^{15}\text{N}$ values for tissues from moderately exposed and sheltered sites in the two study regions ± 1 SD.